

Amendments to the Claims

This listing of claims will replace all prior versions, and listings of claims in the application:

Listing of Claims:

1 (Currently amended). A polynucleotide comprising a sequence encoding a polypeptide that is capable of high level presentation of antigenic peptides on antigen-presenting cells, wherein the polypeptide comprises a β 2-microglobulin molecule that is linked through its carboxyl terminal to a polypeptide stretch that allows the anchorage of the β 2-microglobulin molecule to the cell membrane, and through its amino terminal to at least one antigenic peptide comprising an MHC class I epitope, ~~wherein said antigenic peptide is not related to an autoimmune disease~~ selected from the group consisting of a tumor-associated antigen (TAA), an antigen from a pathogen selected from the group consisting of a bacterial antigen, a viral antigen, a fungal antigen and a parasite antigen, and at least one idiotypic peptide expressed by autoreactive T lymphocytes, wherein said polypeptide stretch at the β ₂-microglobulin carboxyl terminal consists of a bridge peptide which spans the whole distance to the cell membrane, said bridge peptide being linked to a sequence which can exert the required anchoring function, consisting of the full or partial transmembrane and/or cytoplasmic domain of a

molecule selected from the group consisting of the human CD3 ζ polypeptide, CD40 and the MHC I class heavy chain of HLA-A, HLA-B or HLA-C molecule.

Claim 2 (Cancelled).

3(Currently amended). The polynucleotide of claim 1 ~~2~~, wherein said bridge peptide is the peptide of SEQ ID NO: 1, ~~of the sequence: LRWEPSSQPTIPI.~~

Claim 4 (Cancelled).

5(Currently amended). The polynucleotide of claim 4 1, wherein said bridge peptide is linked to the transmembrane and cytoplasmic domains from the MHC class I heavy chain HLA-A2 molecule, ~~of the SEQ ID NO:2, of the sequence:~~
VGIIAGLVLFCAVITCAVVAAVMWRRKSSDRKGCSSYQAASSDSAQCSDEVSLTACKV.

Claims 6-10 (Cancelled).

11(Previously presented). The polynucleotide of claim 1, wherein said at least one antigenic peptide comprising a MHC class I epitope is linked to the β 2-microglobulin amino terminal through a peptide linker.

12(Original). The polynucleotide of claim 1, wherein said at least one antigenic peptide is at least one antigenic determinant of one sole antigen.

13(Original). The polynucleotide of claim 11, wherein said at least one antigenic peptide is at least one antigenic determinant of each one of at least two different antigens.

14(Previously presented). The polynucleotide of claim 12, wherein said antigen is a tumor-associated antigen (TAA).

15(Original). The polynucleotide of claim 14, wherein said TAA is selected from the group consisting of alpha-fetoprotein, BA-46/lactadherin, BAGE, BCR-ABL fusion protein, beta-catenin, CASP-8, CDK4, CEA, CRIPTO-1, elongation factor 2, ETV6-AML1 fusion protein, G250, GAGE, gp100, HER-2/neu, intestinal carboxyl esterase, KIAA0205, MAGE, MART-1/Melan-A, MUC-1, N-ras, p53, PAP, PSA, PSMA, telomerase, TRP-1/gp75, TRP-2, tyrosinase, and uroplakin Ia, Ib, II and III.

16(Previously presented). The polynucleotide of claim 15, wherein said antigenic peptide is selected from the group consisting of:

- (i) the alpha-fetoprotein peptide
GVALQTMKQ (SEQ ID NO:4);
- (ii) the BAGE-1 peptide AARAVFLAL (SEQ ID
NO:5);
- (iii) the BCR-ABL fusion protein peptide
SSKALQRPV (SEQ ID NO:6);

- (iv) the beta-catenin peptide SYLDSGIHF
(SEQ ID NO:7);
- (v) the CDK4 peptide ACDPHSGHFV (SEQ ID
NO:8);
- (vi) the CEA peptide YLSGANLNL (SEQ ID
NO:9);
- (vii) the elongation factor 2 peptide
ETVSEQSNV (SEQ ID NO:10);
- (vii) the ETV6-AML1 fusion protein peptide
RIAECILGM (SEQ ID NO:11);
- (ix) the G250 peptide HLSTAFARV (SEQ ID
NO:12);
- (x) the GAGE-1,2,8 peptide YRPRPRRY (SEQ
ID NO:13);
- (xi) the gp100 peptides KTWGQYWQV (SEQ ID
NO:14), (A)MLGTHTMEV (SEQ ID NO:15),
ITDQVPFSV (SEQ ID NO:16), YLEPGPVTA
(SEQ ID NO:17), LLDGTATLRL (SEQ ID
NO:18), VLYRYGSFSV (SEQ ID NO:19),
SLADTNSLAV (SEQ ID NO:20), RLMKQDFS
(SEQ ID NO:21), RLPRIFCSC (SEQ ID
NO:22), LIYRRRLMK (SEQ ID NO:23),
ALLAVGATK (SEQ ID NO:24), IALNFPQSQK

- (SEQ ID NO:25) and ALNFPGSQK (SEQ ID NO:26);
- (xii) the HER-2/neu peptide KIFGSLAFL (SEQ ID NO:27);
- (xiii) the intestinal carboxyl esterase peptide SPRWWPTCL (SEQ ID NO:28);
- (xiv) the KIAA0205 peptide AEPINIQTW (SEQ ID NO:29);
- (xv) the MAGE-1 peptides EADPTGHSY (SEQ ID NO:30) and SLFRAVITK (SEQ ID NO:31);
- (xvi) the MAGE-3 peptides EVDPIGHLY (SEQ ID NO:32) and FLWGPRALV (SEQ ID NO:33);
- (xvii) the MART-1/Melan-A peptide (E)AAGIGILTV (SEQ ID NO:34);
- (xviii) the MUC-1 peptide STAPPVHNV (SEQ ID NO:35);
- (xix) the N-ras peptide ILDTAGREEY (SEQ ID NO:36);
- (xx) the p53 peptide LLGRNSFEV (SEQ ID NO:37);
- (xxi) the PSA peptides FLTPKKLQCV (SEQ ID NO:38) and VISNDVCAQV (SEQ ID NO:39);
- (xxii) the telomerase peptide ILAKFLHWL (SEQ ID NO:40);

(xxiii) the TRP-1 peptide MSLQRQFLR (SEQ ID NO:41);

(xxiv) the TRP-2 peptides LLGPGRPYR (SEQ ID NO:42), SVYDFFVWL (SEQ ID NO:43), and TLDSQVMSL (SEQ ID NO:44);

(xxv) the TRP2-INT2 peptide EVISCKLIKR (SEQ ID NO:45); and

(xxvi) the tyrosinase peptide KCDICTDEY (SEQ ID NO:46).

17(Previously presented). The polynucleotide of claim 14, wherein said at least one antigenic peptide is at least one antigenic determinant of one sole tumor-associated antigen.

18(Original). The polynucleotide of claim 17, wherein said at least one antigenic peptide is at least one HLA-A2 binding peptide and at least one HLA-A3 binding peptide derived from the melanoma-associated antigen gp100.

19(Original). The polynucleotide of claim 18, wherein said at least one HLA-A2 binding peptide derived from gp100 is selected from the group consisting of SEQ ID NO: 14, 15, 16, 17, 18, 19, 20, 21 and 22, and said at least one gp100 HLA-A3 binding peptide is selected from the group consisting of SEQ ID NO: 23, 24, 25 and 26.

20(Previously presented). The polynucleotide of claim 14, wherein said at least one antigenic peptide is at least one antigenic determinant of each one of at least two different tumor-associated antigens.

21(Original). The polynucleotide of claim 20, wherein said at least one antigenic peptide is at least one HLA-A2 binding peptide derived from each one of the melanoma associated antigens gp100 and Melan-A/MART-1.

22(Original). The polynucleotide of claim 21, wherein said at least one antigenic peptide is at least one HLA-A3-restricted gp100 and at least one HLA-A2-restricted Melan-A/MART-1 peptide.

23(Previously presented). The polynucleotide of claim 12, wherein said antigen is an antigen from a pathogen selected from the group consisting of a bacterial, viral, fungal and parasite antigen.

24(Currently amended). The polynucleotide of claim 23, wherein the antigen is a viral antigen.

25(Currently amended). The polynucleotide of claim 24, wherein the viral antigen is an HIV protein selected from the group consisting of the HIV-1 regulatory proteins Tat and Rev and

the HIV envelope protein, in which case the antigenic peptide derived therefrom has the sequence RGPGRAFVTI (SEQ ID NO:47).

26(Original). The polynucleotide of claim 11, wherein said at least one antigenic peptide is at least one idiotypic peptide expressed by autoreactive T lymphocytes.

27(Original). The polynucleotide of claim 26, wherein said at least one idiotypic peptide is derived from a CDR (complementarity-determining region) sequence of an immunoglobulin or of a TCR chain, optionally containing said CDR flanking segments.

28(Original). The polynucleotide of claim 27, wherein said CDR is the CDR3 of an immunoglobulin or of a TCR chain.

29(Previously presented). The polynucleotide of claim 1 that is an expression vector.

30(Previously presented). An expression vector comprising a polynucleotide according to claim 1.

31(Original). A recombinant viral vector of claim 30.

32(Currently amended). An antigen-presenting cell transfected with a polynucleotide comprising a sequence encoding a polypeptide comprising a β 2-microglobulin molecule that is linked through its carboxyl terminal to a polypeptide stretch

that allows the anchorage of the β_2 -microglobulin molecule to the cell membrane, and through its amino terminal to at least one antigenic peptide comprising a MHC class I epitope, wherein said polypeptide stretch at the β_2 -microglobulin carboxyl terminal consists of a bridge peptide which spans the whole distance to the cell membrane, said bridge peptide being linked to a sequence which can exert the required anchoring function, consisting of the full or partial transmembrane and/or cytoplasmic domain of a molecule selected from the group consisting of the human CD3 ζ polypeptide, CD40 and the MHC I class heavy chain of HLA-A, HLA-B or HLA-C molecule.

33(Original). The antigen-presenting cell of claim 32 selected from the group consisting of a dendritic cell, a macrophage, a B cell and a fibroblast.

34(Currently amended). The antigen-presenting cell of claim 32, wherein said antigenic peptide is a peptide ~~not related to an autoimmune disease~~ comprising a MHC class I epitope selected from a tumor-associated antigen (TAA), an antigen from a pathogen selected from the group consisting of a bacterial antigen, a viral antigen, a fungal antigen and a parasite antigen, and at least one idiotypic peptide expressed by autoreactive T lymphocytes.

35(Original). The antigen-presenting cell of claim 34, wherein said antigenic peptide is at least one peptide derived from at least one TAA.

36(Original). The antigen-presenting cell of claim 34, wherein said antigenic peptide is at least one peptide derived from an antigen from a pathogen selected from the group consisting of a bacterial, a viral, a fungal and a parasite antigen.

37(Previously presented). A DNA vaccine comprising a polynucleotide of claim 1 or an expression vector of claim 30.

38(Currently amended). The DNA vaccine of claim 37 for prevention or treatment of cancer, wherein said polynucleotide is a polynucleotide comprising a sequence encoding a polypeptide that is capable of high level presentation of antigenic peptides on antigen-presenting cells, wherein the polypeptide comprises a β 2-microglobulin molecule that is linked through its carboxyl terminal to a polypeptide stretch that allows the anchorage of the β 2-microglobulin molecule to the cell membrane, and through its amino terminal to at least one antigenic peptide comprising a MHC class I epitope, and said at least one antigenic peptide is at least one antigenic determinant of one sole tumor-associated antigen (TAA).

39(Currently amended). The DNA vaccine of claim 37 for prevention or treatment of a disease caused by a pathogenic organism, wherein said polynucleotide is a polynucleotide comprising a sequence encoding a polypeptide that is capable of high level presentation of antigenic peptides on antigen-presenting cells, wherein the polypeptide comprises a β 2-microglobulin molecule that is linked through its carboxyl terminal to a polypeptide stretch that allows the anchorage of the β 2-microglobulin molecule to the cell membrane, and through its amino terminal to at least one antigenic peptide comprising a MHC class I epitope, and said at least one antigenic peptide is at least one antigenic determinant of one sole antigen from a pathogen selected from the group consisting of a bacterial, viral, fungal and parasite antigen.

40(Original). A cellular vaccine, which comprises an antigen presenting cell of claim 32.

41(Currently amended). The cellular vaccine of claim 40, wherein the antigen presenting cell is selected from the group consisting of a dendritic cell, a macrophage, a B cell and a fibroblast.

42(Currently amended). The cellular vaccine of claim 41, wherein the at least one antigenic peptide presented by the

antigen presenting cell is a peptide ~~not related to an autoimmune disease~~ comprising a MHC class I epitope from a tumor-associated antigen (TAA), an antigen from a pathogen selected from the group consisting of a bacterial antigen, a viral antigen, a fungal antigen and a parasite antigen, and at least one idiotypic peptide expressed by autoreactive T lymphocytes.

43(Currently amended). The cellular vaccine of claim 42 for prevention or treatment of cancer, wherein the antigen presenting cell presents at least one peptide derived from at least one tumor associated antigen.

44(Currently amended). The cellular vaccine of claim 42 for prevention or treatment of a disease caused by a pathogenic organism, wherein the antigen presenting cell presents at least one peptide derived from a pathogenic antigen.

Claims 45-46 (cancelled).

47(Currently amended). A method of immunizing a mammal against a tumor-associated antigen comprising the step of immunizing the mammal with a cellular vaccine, which comprises an antigen presenting cell transfected with a polynucleotide comprising a sequence encoding a polypeptide comprising a β 2-microglobulin molecule that is linked through its carboxyl terminal to a polypeptide stretch that allows the anchorage of

the β 2-microglobulin molecule to the cell membrane, and through its amino terminal to at least one antigenic peptide comprising a MHC class I epitope, wherein said antigen presenting cell is selected from the group consisting of a dendritic cell, a macrophage, a B cell or a fibroblast, and said at least one antigenic peptide is at least one peptide derived from at least one tumor-associated antigen, wherein said polypeptide stretch at the β 2-microglobulin carboxyl terminal consists of a bridge peptide which spans the whole distance to the cell membrane, said bridge peptide being linked to a sequence which can exert the required anchoring function, consisting of the full or partial transmembrane and/or cytoplasmic domain of a molecule selected from the group consisting of the human CD3 ζ polypeptide, CD40 and the MHC I class heavy chain of HLA-A, HLA-B or HLA-C molecule.

48(Currently amended). A method of immunizing a mammal against a disease caused by a pathogenic organism comprising the step of immunizing the mammal with a cellular vaccine, which comprises an antigen presenting cell transfected with a polynucleotide comprising a sequence encoding a polypeptide comprising a β 2-microglobulin molecule that is linked through its carboxyl terminal to a polypeptide stretch that allows the anchorage of the β 2-microglobulin molecule to the cell membrane, and through its amino terminal to at least one antigenic peptide

comprising a MHC class I epitope, wherein said antigen presenting cell is selected from the group consisting of a dendritic cell, a macrophage, a B cell or a fibroblast, and said at least one antigenic peptide is at least one antigenic peptide derived from a pathogen selected from the group consisting of a bacterial antigen, a viral antigen, a fungal antigen and a parasite antigen, wherein said polypeptide stretch at the β_2 -microglobulin carboxyl terminal consists of a bridge peptide which spans the whole distance to the cell membrane, said bridge peptide being linked to a sequence which can exert the required anchoring function, consisting of the full or partial transmembrane and/or cytoplasmic domain of a molecule selected from the group consisting of: the human CD3 ζ polypeptide, CD40 and the MHC I class heavy chain of HLA-A, HLA-B or HLA-C molecule.

49(Currently amended). A pharmaceutical composition, comprising as an active ingredient at least one polynucleotide of claim 1 and a pharmaceutically acceptable carrier.

50(Currently amended). The pharmaceutical composition of claim 49, wherein the polynucleotide comprises a sequence encoding a polypeptide comprising at least one antigenic peptide derived from at least one tumor associated antigen.

51(Currently amended). The pharmaceutical composition of claim 49, wherein the polynucleotide comprises a sequence

encoding a polypeptide comprising at least one antigenic peptide derived from a pathogenic antigen.

52 (Currently amended). A pharmaceutical composition, comprising as an active ingredient at least one antigen presenting cell of claim 32 and a pharmaceutically acceptable carrier.

53 (Previously presented). The polynucleotide of claim 13, wherein said antigen is a tumor-associated antigen (TAA).

Claims 54 and 55 (Cancelled).

56 (New). A polynucleotide comprising a sequence encoding a polypeptide that is capable of high level presentation of antigenic peptides on antigen-presenting cells, wherein the polypeptide comprises a β 2-microglobulin molecule that is linked through its carboxyl terminal to a polypeptide stretch that allows the anchorage of the β 2-microglobulin molecule to the cell membrane and through its amino terminal to at least one antigenic peptide comprising a MHC class I epitope selected from the group of peptides consisting of SEQ ID NOs:4 to 46, wherein said polypeptide stretch at the β 2-microglobulin carboxyl terminal consists of a bridge peptide which spans the whole distance to the cell membrane, said bridge peptide being linked to a sequence which can exert the required anchoring function, consisting of

the full or partial transmembrane and/or cytoplasmic domain of a molecule selected from the group consisting of the human CD3 ζ polypeptide and the MHC I class heavy chain of HLA-A2 of SEQ ID NO: 2.

57(New). A polynucleotide comprising a sequence encoding a polypeptide that is capable of high level presentation of antigenic peptides on antigen-presenting cells, wherein the polypeptide comprises a β_2 -microglobulin molecule that is linked through its carboxyl terminal to a polypeptide stretch that allows the anchorage of the β_2 -microglobulin molecule to the cell membrane and through its amino terminal to at least one antigenic peptide comprising a MHC class I epitope selected from the group of peptides consisting of SEQ ID NOs:4 to 47, wherein said polypeptide stretch at the β_2 -microglobulin carboxyl terminal consists of a bridge peptide which spans the whole distance to the cell membrane, said bridge peptide being linked to a sequence which can exert the required anchoring function, consisting of the full or partial transmembrane and/or cytoplasmic domain of a molecule selected from the group consisting of the human CD3 ζ polypeptide, the MHC I class heavy chain of HLA-A2 of SEQ ID NO: 2 and CD40.